

The NIH Pain Consortium
3rd Annual Symposium on Advances in Pain Research
Natcher Auditorium/NIH Campus
May 22, 2008

Welcome and Introduction

The meeting was opened by Dr. Linda Porter, Ph.D., Program Director for pain research at the National Institute for Neurological Disorders and Stroke.

Opening Remarks: NIH Pain Consortium Co-Chair

Patricia A. Grady, Ph.D., R.N., Director of the National Institute for Nursing Research

Dr. Grady spoke on behalf of the NIH Pain Consortium, and noted that she was representing not only herself but also Dr. Story Landis, Director of the National Institute for Neurological Disorders and Stroke, Dr. Larry Tabak, Director of the National Institute of Dental and Craniofacial Research, and all of the 20 NIH Institutes and Centers participating in the Consortium. She said that this year's level of NIH funding for pain research is estimated to be 222 million dollars, and funding is currently at steady state. The Consortium is aware that this funding level is modest relative to the overall problem of pain.

She explained that the mission of the Pain Consortium (founded in 1996) is to increase the visibility of and develop a comprehensive NIH agenda for pain research, to support and promote interdisciplinary efforts in this area, and to pursue public-private partnerships. Consortium activities include identifying research gaps and opportunities, and coordinating the release of funding initiatives to address them. One area in which there has been particular progress recently is the genetics of pain, and advances in this area are opening up new prospects for individualized medicine. Emotional and bio-behavioral aspects of pain are also receiving increasing attention, and more sophisticated tools for their study are now becoming available. Dr. Grady presented a list of current NIH funding opportunities in pain research, most of which represent new initiatives. She closed by applauding the commitment of pain researchers and advocates to improving the quality of life of those suffering from pain.

Opening Remarks: American Pain Society Representative

Jennifer Haythornthwaite, Ph.D., Johns Hopkins University

On behalf of the American Pain Society, Dr. Haythornthwaite presented epidemiological data on the prevalence of pain in the US and the world. International studies estimate that 11 percent of the general adult population reports chronic pain, and 22 percent of patients seeking primary care report pain that has lasted longer than six months. The percentages of individuals reporting pain and pain-related activity limitations are higher among older adults, and with the aging of the population we are facing a growing pain epidemic. Another cause for concern with respect to the older population comes from a recent study showing high rates of persistent pain following various surgical procedures, including hernia repair and coronary bypass surgery. Pain impacts not only subjective

well-being, but day-to-day functioning, sleep, mood, and social functioning. Inadequate pain control may also contribute to disease progression. Pain is also a very expensive problem in terms of dollars spent on health care, disability, and absenteeism.

Dr. Haythornthwaite described the mission and activities of the American Pain Society, and pointed out that the years 2000 to 2010 have been named the Decade of Pain Research. She listed among current research challenges the development of better preclinical models, the exploration of new mechanisms of pain, and faster translation from the bench to clinical studies and clinical care.

PANEL SESSION

Basic Mechanisms Underlying Pain Therapies

Moderator: John Kusiak, Ph.D. National Institute of Dental and Craniofacial Research

Cannabinoid Analgesia Through TRPV1 Desensitization

Ken Hargreaves, D.D.S., Ph.D., University of Texas Health Science Center

Cannabinoids (CBs) are a family of bioactive compounds originally extracted from the plant *Cannabis sativa* (marijuana). Certain groups of nerve cells in the human brain produce endocannabinoids, molecules with different structures but with activity similar to the plant derived compounds. Previous research has shown that CBs have anti-hyperalgesic activity in animal models of inflammatory pain, but the mechanism of this effect has been mysterious. Recent work from Dr. Hargreaves' laboratory indicates that this anti-pain effect of CBs is mediated by transient receptor potential vanilloid type 1 (TRPV1) receptors. TRPV1 receptors are found on a large subpopulation of peripheral nociceptive (pain-sensing) neurons, and are activated by painful stimuli such as capsaicin (the molecule that makes hot peppers hot). CBs also interact with these receptors: if a CB-like drug is applied to neurons shortly before a painful stimulus, the TRPV1 receptor response to the painful stimulus is greatly reduced. This phenomenon, called "receptor desensitization," suggests a mechanism whereby CBs could exert their anti-hyperalgesic effects.

CBs can also desensitize a second class of TRP receptors on nociceptive neurons, TRPA1 receptors. Moreover, work from the laboratory of Dr. Ardem Akopian showed that subunits of the TRPV1 and TRPA1 receptors can bind to one another to form a novel form of the TRP1 receptor, called a heterodimer. This heterodimeric receptor represents a new therapeutic target for the development of CBs that would be devoid of classic CB-like side effects. The heterodimeric TRP1 receptor also represents a novel class of receptors for CBs, which were previously thought to exert their biological effects entirely via CB1 and CB2 receptors.

Cannabinoid and Sphingolipid Enhancement of Opioid Antinociception

Sandra Welch, Ph.D., Virginia Commonwealth University

Cannabinoids (CBs) such as THC not only have analgesic activity of their own, but also enhance the analgesic effects of opioids. This enhancement occurs even at very low CB doses, including doses that have little or no analgesic activity when the CB is administered alone. For example, orally administered THC produces a two-fold increase in the potency of morphine, and a greater than 25-fold increase in the potency of codeine. Thus, by administering a CB together with an opioid, one can greatly reduce the dose of opioid needed to manage pain, and also reduce opioid side effects. Co-administration of a CB can also prevent the development of opioid tolerance and physical dependence. More recently, Dr. Welch has shown that opioid activity is also enhanced by co-administration of a CB that is produced naturally in the brain, anandamide. To achieve this effect, anandamide was administered together with another drug (URB-57) that inhibits the breakdown of anandamide by one of the body's enzymes.

Dr. Welch's group also has been studying the potential use of sphingosine-1-phosphate (S1P) to enhance opioid effects. S1P is a lipid (fat) molecule that is produced by many different cell types and functions as a cell-signaling molecule. S1P regulates a variety of cellular processes via activation of a specific family of receptors (S1P receptors). S1P receptors and CB receptors show very similar patterns of distribution in the brain, suggesting that these two receptor systems might interact with each other. In addition, S1P produces biological effects similar to those of the CBs.

Migraine and Cortical Spreading Depression: Effects on the Brain and How We Should Treat It

Maiken Nedergaard, M.D., Ph.D., University of Rochester Medical Center

Migraine is one of the most common neurological conditions associated with pain. Migraine differs however, from certain other pain syndromes in that its clinical management must be geared not toward treating the symptom of pain *per se*, but rather preventing it in the first place by treating the underlying disease. Much current research on the mechanisms of migraine is focused on cortical spreading depression (CSD), a neurological phenomenon often associated with migraine. CSD is a brief wave of neuronal excitation that moves across the cortex, and is immediately followed by a prolonged period of neuronal depression, together with severe constriction of blood vessels.

CSD is accompanied by large, widespread movements of ions (charged particles) in and out of brain cells. Returning these ions to their original states of balance requires the large-scale activation of protein pumps in cell membranes, a process that in turn is likely to require enormous amounts of cellular energy. To test this idea, Dr. Nedergaard's laboratory used advanced optical imaging techniques to visualize the levels and cellular distribution of NADH during CSD in mice. NADH is a molecule used in cellular energy metabolism, and its production increases under conditions of high oxygen demand. During CSD, large increases in NADH levels are seen in neurons that lie in the areas between blood vessels. These neurons usually receive less oxygen than those lying close to blood vessels. Thus, it seems probable that these neurons become oxygen-deprived during CSD, with adverse consequences for their function. In support of that idea, the

neurons can be seen to undergo severe swelling during CSD, and their dendrites (cellular processes that receive input from other neurons) lose their normal structure. These observations highlight the importance of preventing episodes of migraine as well as treating the pain associated with them.

Mechanisms of Triptan Action in Migraine and Pain

Andrew Ahn, M.D., Ph.D., University of California San Francisco

Triptans are drugs that bind to a subgroup of receptors for the neurotransmitter serotonin (5-HT). These drugs show remarkable clinical specificity in that they are highly effective for treating migraine and cluster headache, but have no effect on other types of pain. To better understand the mechanisms of triptan action in migraine, Dr. Ahn has studied the distribution and regulation of 5-HT_{1D} receptors, a subgroup of the 5-HT receptors to which triptans bind. These receptors are found on subpopulations of peripheral sensory neurons, including those responsible for migraine pain. More specifically, 5-HT_{1D} receptors are found within the endings of sensory nerve processes that project to the spinal cord, where presumably they are involved in the central processing of pain signals. It is puzzling however, that the receptors are not displayed on the cell surface as most receptors are, but rather are sequestered inside vesicles (intracellular storage compartments within the nerve ending). It was unclear therefore as to how neurotransmitters or drugs could interact with these receptors. Dr. Ahn and co-workers showed that sensory neurons actually redistribute their 5-HT_{1D} receptors following a painful stimulus, moving them out from vesicles and onto the surface membrane of the nerve ending, where they are then available to bind 5-HT or drugs like the triptans.

5-HT_{1D} receptors are found not only on the endings of peripheral sensory neurons, but also on nerve endings within in the amygdala, a brain region involved in emotional processing. Hence, this population of 5-HT_{1D} receptors may contribute to emotional components of pain, and to reported anti-anxiety effects of the triptans.

PANEL SESSION

Basic Discoveries: Moving Towards Clinical Care

Moderator: *Wen Chen, Ph.D., National Institute on Aging*

Selective Targeting of Pain Fibers in Regional Anesthesia

Bruce Bean, Ph.D., Harvard Medical School

Sodium channels are neuronal cell membrane proteins, each of which contains a central pore that allows sodium and other ions (charged particles) to pass from the outside to the inside of the cell. Movement of sodium ions through these channels in turn generates action potentials, the electrical impulses by which neurons communicate. Lidocaine and other local anesthetics work by blocking sodium channels so that ions can no longer pass through them, thereby blocking the generation of nerve impulses. Lidocaine however, blocks sodium channels in motor and autonomic neurons as well as sensory ones, and thus produces undesirable side effects.

Dr. Bean and colleagues found it is possible to block sodium channels selectively in sensory neurons using a drug called QX314. This drug is structurally related to lidocaine, but does not block sodium channels when applied to the outside of a cell. Rather, QX314 first has to enter the cell by passing through other channels in the cell membrane, and then binds to sodium channels from the inside. One kind of channel through which QX314 may be able to enter cells is the TRPV1 channel, which has a relatively large pore. In addition, this channel is found only on sensory neurons, suggesting it could serve as a conduit for targeting QX314 specifically into sensory neurons. And indeed, when QX314 is applied to sensory neurons in culture together with a drug that opens TRPV1 channels (capsaicin), it produces a long-lasting inhibition of neuronal firing. Co-application of QX314 and capsaicin also produces long-lasting analgesic effects in a rat model, and without the motor side effects normally seen with lidocaine. Because capsaicin initially causes pain in this regimen however, Dr. Bean and colleagues have been looking for alternative drugs that open TRPV1 channels and can be co-administered with QX314. They have now identified at least one such drug: lidocaine itself. Lidocaine binds to TRPV1 channels as well as sodium channels, and when applied briefly allows QX314 to enter sensory neurons via TRPV1 channels and inhibit neuronal firing for prolonged periods. This drug co-administration strategy could prove particularly useful for preventing post-operative pain.

Improgan: A Cannabinomimetic Analgesic
Lindsay Hough, Ph.D., Albany Medical School

Improgan is a drug structurally related to histamine, which functions as both an immune regulator and a neurotransmitter. When injected into the brain, improgan has a broad range of anti-nociceptive (anti-pain) effects. Improgan does not appear to act via opioid receptors. It may act in part through activation of the brain's endogenous cannabinoid pathways, but evidence for this mechanism remains incomplete. Dr. Hough and colleagues recently identified another potential improgan target: the cytochrome P450 (CYP) family of proteins. CYPs are enzymes that catalyze a wide variety of metabolic reactions inside the body, including the breakdown of food substances and drugs and the synthesis of endogenous biological compounds such as hormones and cholesterol. A drug that blocks the anti-nociceptive effect of improgan was found to inhibit CYPs, and mice whose CYP levels are reduced by genetic modification show reduced improgan anti-nociception. Preliminary results suggest that improgan acts via the CYP system to generate epoxyeicosatrienoic acids (EETs). EETs are fatty acids that act as signaling molecules, and whose effects include relaxation of blood vessels and reduction of inflammation. Together, these data point to the EETs as a new group of potential targets for therapeutics development.

Intrathecal AAV: A Candidate Platform for Translational Pain Research
Andreas Beutler, M.D., Mount Sinai School of Medicine

Gene therapy offers several important advantages over traditional drug therapy. These include the ability to target specific anatomical sites or cell populations, and to obtain

long-lasting therapeutic effects with a single administration. Gene therapy in the nervous system poses special challenges, however. Most gene therapy delivery systems, or “vectors” (usually viruses), don’t cross the blood-brain barrier and so must be injected directly into neural tissue. In addition, viral vectors often fail to infect neural cells at the levels necessary to obtain useful levels of therapeutic gene expression.

Dr. Beutler and colleagues have overcome these technical obstacles by (1) injecting viral vectors by lumbar puncture into the narrow, fluid-filled space that surrounds the spinal cord, and (2) identifying a particular species of adeno-associated virus (AAV) that efficiently infects axons in the spinal cord, including those of peripheral nociceptive neurons. The vector is transported out to the cell bodies of these neurons (which lie outside the spinal cord) and can replicate inside them to generate large numbers of copies of a therapeutic gene. Using this system, a gene for an analgesic opioid peptide was delivered into an animal model of neuropathic pain. Reduction in pain responses were seen within one month of gene vector administration, and this therapeutic effect persisted for at least four months. In his discussion of these results, Dr. Beutler emphasized that the production of sufficient quantities of high quality gene therapy vectors for use in clinical trials is now a realistic goal, and one that should be pursued actively.

Matrix Metalloproteinases: A New Target for Pain Therapy?

Ru-Rong Ji, Ph.D., Brigham and Women’s Hospital

Matrix metalloproteinases (MMPs) are a large family of enzymes thought to play a central role in the development of inflammatory responses and associated pain after tissue injury. Dr. Ji and colleagues have identified two MMPs in particular as critical mediators of neuropathic pain: MMP-9 and MMP-2. Both enzymes appear to act in part by activating interleukin-1B (IL-1B), an intercellular signaling protein that promotes pain sensitivity. The two enzymes play different roles in the development of pain sensitivity after injury, however. First, they have different time courses of action: MMP-9 levels rise transiently for the first few days following injury, whereas increases in MMP-2 follow later and persist for weeks. Second, they are produced by different cell types: MMP-9 is made by peripheral sensory neurons, whereas MMP-2 is generated by surrounding glial cells (cells that support and regulate neuronal cell function). Finally, the two MMPs differ in their downstream actions: MMP-9 activates IL-1B in microglial cells (circulating cells which arise from the immune system), whereas MMP-2 activates IL-1B in astrocytes (glial cells that permanently reside together with neurons in sensory ganglia).

Drugs that inhibit MMP-9 or MMP-2 caused dramatic reductions in pain sensitivity in a rodent model of neuropathic pain. MMP-9 inhibitors however, were effective only at early time points after injury and MMP-2 inhibitors only at later time points. These results are consistent with the differential temporal profiles of expression of these two enzymes after injury, and suggest that different treatments should be considered at different stages of chronic pain development.

PANEL SESSION

Parallel Pathways: Basic and Clinical Discoveries

Moderator: Kathy Mann Koepke, Ph.D., National Institute of Nursing Research

Complex Genetics of Pain Susceptibility

William Maixner, D.D.S., Ph.D., University of North Carolina School of Dentistry

Dr. Maixner's research has focused on genetic risk factors that contribute to the development of chronic pain conditions. Studies in both humans and animal models indicate that genetic factors account for approximately 50% of the variation in pain sensitivity seen among different individuals. Dr. Maixner and coworkers found that an individual's level of pre-existing pain sensitivity contributes to that person's risk of developing chronic pain conditions such as temporomandibular joint disorder (TMJD). A search for specific genetic differences between low, average, and high pain sensitivity individuals pointed to the gene for catecholamine-O-methyl transferase (COMT). COMT is an enzyme involved in the breakdown of the neurotransmitters known as catecholamines (norepinephrine, epinephrine, and dopamine). Variations in this gene also were associated with the likelihood of developing TMJD, as were variations in the gene for the beta-2 adrenergic receptor (one of the receptors to which catecholamines bind). Cell culture studies showed that the COMT gene variant associated with high pain sensitivity caused reduced cellular production of the enzyme.

In further support of the hypothesis that COMT is involved in pain sensitivity, an inhibitor of this enzyme was found to increase pain sensitivity in rodent models. The COMT inhibitor also increased levels of cytokines which are molecules known to be involved in pain and inflammation. The increases in pain sensitivity and cytokine levels both were reversed by drugs that block beta-2 or beta-3 adrenergic receptors. Dr. Maixner and colleagues are currently undertaking a large-scale (3200 individuals) prospective study to identify additional risk factors for TMJD, including psychosocial as well as genetic factors.

Genetic Variation in NPY Alters Human Pain/Stress Response and Emotion

David Goldman, M.D., National Institute on Alcohol Abuse and Alcoholism

Neuropeptide Y (NPY) is a peptide (small protein) neurotransmitter that is released during stress and helps mediate the body's recovery from stress. NPY is of particular interest to pain researchers because it can reduce both pain sensation and anxiety. Hence, NPY may be involved in modulating emotional responses to pain, and also increases in pain sensitivity that accompany negative emotional states. Dr. Goldman and colleagues have identified variations in the NPY gene that result in relatively low or high production of the peptide (as measured, for example, by checking blood NPY levels). Individuals carrying the low production NPY gene variant show more pronounced activation of the amygdala (a brain region involved in fear responses) when presented with negative emotional stimuli than do individuals carrying the high production NPY gene variant. They also showed diminished resilience to pain and stress as assessed by activation of endogenous opioid neurotransmission in various brain regions. The gene variation responsible for the differences in NPY production lies in a single nucleotide (or "letter")

in the DNA code) in the region of the gene that controls the rate at which the gene is transcribed to produce mRNA . These differences in the level of mRNA lead to the differences in the levels of NPY peptide.

Quality of Life and Pain in Premenopausal Women with Major Depressive Disorder
Ann Berger, M.D., M.S.N., NIH Clinical Center

Pain and depression are closely intertwined conditions, and both are associated with sleep problems and impaired quality of life. To better quantify the relationship between pain and depression, Dr. Berger and coworkers studied the prevalence of pain in premenopausal women with major depressive disorder (MDD). Compared to healthy controls, women with MDD reported head and neck pain at much higher rates (48% of women with MDD versus 16% of controls), although the average intensity of the pain they experienced was relatively mild. The elevated rates of pain in women with MDD were associated with increased fatigue, anxiety, concentration and memory problems, and insomnia. Women with MDD also experienced a lower quality of life, especially in the psychological domain. For example, they complained more of negative daily stressors, including worries about physical appearance, misplacing things, and not having enough energy. It is interesting that women with MDD also had higher circulating levels of two pain-related neuropeptides, substance P and calcitonin gene-related peptide. An outstanding question remains as to whether elevated pain levels in this population lead to increased depressive symptoms, or whether depression leads to increased pain.

Behavioral and Neural Underpinnings of Pain Processing in Major Depressive Disorder
Irina Strigo, Ph.D., University of California San Diego

Pain is both a sensation and an emotional experience, and pain and depression often occur together. To further investigate the relationship between depression and pain, Dr. Strigo and coworkers assessed emotional responses to heat stimuli of increasing intensities in young adults with Major Depressive Disorder (MDD). Compared to healthy controls, MDD subjects showed decreased heat pain thresholds (they were more sensitive to painful heat). In addition, they showed increased negative emotional responses in terms of their individual experiences of the unpleasantness and/or intensity of the stimuli. For example, some MDD patients would rate a warm stimulus as painful and unpleasant, when controls rated the same stimulus as non-painful and even pleasant.

Dr. Strigo and coworkers found that MDD subjects also differ from controls in the extent to which certain regions of their brains are activated during pain anticipation and pain experience. During pain anticipation, MDD subjects showed increased activation of the amygdala, insula, and anterior cingulate cortex (all brain regions that are involved in emotional processing). During pain experience, MDD subjects showed decreased activation in the anterior cingulate and the periaqueductal gray (another region involved in endogenous pain modulation).